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(54) Title: DSRNA AS INSECT CONTROL AGENT

(57) Abstract: The present invention relates to methods for controlling pest infestation using double stranded RNA molecules. The invention provides methods for making transgenic plants that express the double stranded RNA molecules, as well as pesticidal agents and commodity products produced by the inventive plants.

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INTERNATIONAL SEARCH REPORT

International application No

PCT/IB2006/004003

A. CLASSIFICATION OF SUBJECT MATTER

INV. C12N15/82 A01H5/00 C12N5/10 C12N15/12 C12N15/11

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

C12N A01H C07K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, PAJ, Sequence Search, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	DATABASE UniProt [Online] 30 August 2005 (2005-08-30), "Ribosomal protein S4e." XP002432593 retrieved from EBI accession no. UNIPROT:Q4GXU7 Database accession no. Q4GXU7 abstract	2,3,6
X	& DATABASE EMBL SEQUENCE LIBRARY [Online] Ebi. hinxton; ribosomal protein S4e; rpS4e gene 16 July 2005 (2005-07-16), LONGHORN,S.J.: "Biphyllus lunatus mRNA for ribosomal protein S4e" retrieved from EBI. HINXTON accession no. www.ebi.co.uk Database accession no. AM048926 abstract	2,3,6
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☒ Further documents are listed in the continuation of Box C.

☒ See patent family annex.

* Special categories of cited documents :

- *A* document defining the general state of the art which is not considered to be of particular relevance
- *E* earlier document but published on or after the international filing date
- *L* document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- *O* document referring to an oral disclosure, use, exhibition or other means
- *P* document published prior to the international filing date but later than the priority date claimed

T later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

X document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

Y document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

Z document member of the same patent family

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C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 2005/019408 A (BAR ILAN UNIVERSITY; HAZERA GENETICS LTD; RAHAN MERISTEM LTD; MICHAEL) 3 March 2005 (2005-03-03) the whole document	18
X	WO 2005/049841 A (COMMONWEALTH SCIENTIFIC AND INDUSTRIAL RESEARCH ORGANISATION; BAYER BI) 2 June 2005 (2005-06-02) page 23	18
A	WO 01/34815 A (CAMBRIA BIOSCIENCES, LLC) 17 May 2001 (2001-05-17) the whole document	
A	WO 01/37654 A (DNA PLANT TECHNOLOGY CORPORATION; TOBIAS, CHRISTIAN; SHAH, GOWRI; GUTT) 31 May 2001 (2001-05-31) the whole document	
A	WO 03/004644 A (COMMONWEALTH SCIENTIFIC AND INDUSTRIAL RESEARCH ORGANISATION; WHYARD,) 16 January 2003 (2003-01-16) the whole document claim 13 page 23 - page 24 page 20, line 21	
A	WO 02/46432 A (AVENTIS CROPS SCIENCE GMBH; GUNKEL, NIKOLAS) 13 June 2002 (2002-06-13) the whole document	
A	WO 01/09301 A (GENOPTERA LLC [US]) 8 February 2001 (2001-02-08) the whole document	
A	WO 2004/001000 A (MONSANTO TECHNOLOGY LLC [US]) 31 December 2003 (2003-12-31) page 16, line 21 - page 17; line 2; claim 16	
P,X	WO 2005/110068 A (MONSANTO TECHNOLOGY LLC; BAUM, JAMES, A; GILBERTSON, LARRY, A; KOVALIC) 24 November 2005 (2005-11-24) page 5, line 7 claim 39; example 6	18,27

INTERNATIONAL SEARCH REPORT

International application No.
PCT/IB2006/004003

Box No. II Observations where certain claims were found unsearchable (Continuation of Item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. ☐ As all searchable claims could be searched without effort justifying an additional fees, this Authority did not invite payment of additional fees.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☒ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Invention 1: 1-11, 18-19, 23-25, 27, 38-40 partially

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.
- ☐ The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.
- ☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

Invention 1: 1-11,18-19,23-25,27,38-40 partially

Isolated Leptinotarsa-specific nucleotide sequence as characterized by SEQID1; double stranded ribonucleotide sequence (dsRNA) produced by expressing said SEQID1; cell or plant transformed by said sequence, a product produced from said plant, a method for controlling or preventing insect growth comprising providing a pest with a plant material comprising said dsRNA molecule method for improving crop yield comprising introducing said nucleotide sequence into a plant wherein the expression of said polynucleotide inhibits pest infestation and loss of yield; use of said polynucleotide sequence, said dsRNA, said plant for treating insect, nematode or fungal infestation in plants.

Inventions 2-149: claims 1-11,18-19,23-25,27,38-40 partially

as invention 1, but limited to the Leptinotarsa-specific SEQIDs 3, 5, 7, 9, 11, 13, 15, 17, 19, 21, 23, 49 to 158, 159, 160-163, 168, 173, 178, 183, 188, 193, 198, 203, 208, 215, 220, 225, 230, 240 to 246 and 2486.

Invention 150: claims 1-11,18-19,23-25,38-40 partially, 28 completely

as invention 1, but limited to the Phaendon-specific nucleotide sequences as characterized by SEQIDs 247, 249, 251, 253, 255, 257, 259, 275 to 472, 473, 478, 483, 488, 493, 498, 503, 508 to 512 and the use for Phaendon-specific infestation in plants.

Invention 151: claims 1-11,18-19,23-25,38-40 partially, 29 completely

as invention 1, but limited to the Epilachna-specific nucleotide sequences as characterized by SEQIDs 513, 515, 517, 519, 521, 533 to 575, 576, 581, 586, 591 or 596 and the use for Epilachna-specific infestation in plants.

Invention 152: claims 1-11,18-19,23-25,38-40 partially, 30 completely

as invention 1, but limited to the Anthonomus-specific nucleotide sequences as characterized by SEQIDs 601, 603, 605, 607, 609, 621 to 767, 768, 773, 778, 783 or 788 and the use for Anthonomus-specific infestation in plants.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Invention 153: claims 1-11,18-19,23-25,38-40 partially, 31 completely

as invention 1, but limited to the Tribolium-specific nucleotide sequences as characterized by SEQIDs 793, 795, 797, 799, 801, 813 to 862, 863, 868, 873, 878 or 883 and the use for Tribolium-specific infestations.

Invention 154: claims 1-11,18-19,23-25,38-40 partially, 32 completely

as invention 1, but limited to the Myzus-specific nucleotide sequences as characterized by SEQIDs 888, 890, 892, 894, 896, 908 to 1040, 1041, 1046, 1051, 1056, 1061, or 1066 to 1070 and the use for Myzus-specific infestation in plants.

Invention 155: claims 1-11,18-19,23-25,38-40 partially, 33 completely

as invention 1, but limited to the Nilaparvata-specific nucleotide sequences as characterized by SEQIDs 1071, 1073, 1075, 1077, 1079, 1081, 1083, 1085, 1087, 1089, 1091, 1093, 1095, 1097, 1099, 1101, 1103, 1105, 1107, 1109, 1111, 1113, 1161 to 1571, 1572, 1577, 1582, 1587, 1592, 1597, 1602, 1607, 1612, 1617, 1622, 1627, 1632, 1637, 1642, 1647, 1652, 1657, 1662, 1667, 1672 or 1677 and the use for Nilaparvata-specific infestations in plants.

Invention 156: claims 1-11,18-19,23-25,38-40 partially, 34 completely

as invention 1, but limited to Chilo-specific nucleotide sequences as characterized by SEQIDs 1682, 1684, 1686, 1688, 1690, 1692, 1694, 1696, 1698, 1700, 1702, 1704, 1730 to 2039, 2040, 2045, 2050, 2055, 2060, 2065, 2070, 2075, 2080, 2085, 2090 or 2095 and the use for Chilo-specific infestation in plants.

Invention 157: claims 1-11,18-19,23-25,38-40 partially, 35 completely

as invention 1, but limited to Plutella-specific nucleotide sequences as characterized by SEQIDs 2100, 2102, 2104, 2106, 2108, 2120 to 2338, 2339, 2344, 2349, 2354, or 2359 and the use for Plutella-specific infestations in plants.

Invention 158: claims 1-11,18-19,23-25,38-40 partially, 36 completely

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

as invention 1, but limited to Acheta-specific nucleotide sequences as characterized by SEQIDs 2364, 2366, 2368, 2370, 2372, 2384 to 2460, 2461, 2466, 2471, 2476 or 2481 and the use for Acheta-specific infestations.

Invention 159: claims 12-17 completely

Plant comprising dsRNA derived from a pest species, wherein said dsRNA inhibits pest biological activity or expression of a target sequence, and wherein said target sequence is an insect, nematode, bacteria or fungi sequence, and wherein said plant is further male-sterile.

Invention 160: claim 20

Pesticide comprising a plant expressing a target polynucleotide sequence.

Invention 161: claims 21,22,26 completely

Method for controlling pest infestation comprising identifying a target sequence in a pest, introducing said sequence or an orthologous sequence of a second pest into said plant.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No

PCT/IB2006/004003

Patent document cited in search report		Publication date	Patent family member(s)	Publication date
WO 2005019408	A	03-03-2005	AU 2004267254 A1 CA 2535132 A1 EP 1656012 A2 US 2007199100 A1	03-03-2005 03-03-2005 17-05-2006 23-08-2007
WO 2005049841	A	02-06-2005	EP 1687435 A1	09-08-2006
WO 0134815	A	17-05-2001	AU 1461701 A CA 2388753 A1 EP 1230376 A1 US 6326193 B1 US 2002137710 A1	06-06-2001 17-05-2001 14-08-2002 04-12-2001 26-09-2002
WO 0137654	A	31-05-2001	AU 2047001 A	04-06-2001
WO 03004644	A	16-01-2003	CA 2455490 A1 EP 1414959 A1 JP 2004532653 T NZ 530969 A US 2005095199 A1	16-01-2003 06-05-2004 28-10-2004 30-09-2005 05-05-2005
WO 0246432	A	13-06-2002	AU 1914002 A EP 1210875 A1	18-06-2002 05-06-2002
WO 0109301	A	08-02-2001	AU 6382400 A EP 1206537 A2	19-02-2001 22-05-2002
WO 2004001000	A	31-12-2003	AU 2003251579 A1 EP 1516051 A2	06-01-2004 23-03-2005
WO 2005110068	A	24-11-2005	AU 2005244258 A1 BR PI0509743 A CA 2562022 A1 EP 1732379 A2	24-11-2005 25-09-2007 24-11-2005 20-12-2006